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Evaluation of a universal selective *Burkholderia* enrichment medium with potential use in CF patients

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Introduction and aims: In CF patients respiratory colonisation with *Burkholderia* (B) is associated with poor prognosis. Therefore appropriate microbiological detection of B is important. Several selective agars and enrichment media for B have been described. Since CF is a relatively rare disease, these special media are not available in most clinical laboratories. The aim of this study is to evaluate the potential use of a selective B enrichment medium (SBEM) available to all laboratories.
Methods: The SBEM consists of 2 ml tryptic soy broth supplemented with 2 antibiotic disks: vancomycine (70 μg) and polymyxin B (150 μg) (Neosensitabs, Rosco, Denmark).A volume of 0.5 ml sputum or strain culture is added to SBEM and incubated overnight at 37°C. CF isolates of B representing the most important species involved in CF were selected (n=20) and mixed with bacterial species typically colonizing the respiratory tract of CF patients. Bacterial suspensions of 0.5 Mc Farland were used to evaluate the selectivity and sensitivity of SBEM in non-B/B ratios of 1/1 to 1/1000. CF sputum samples (n=5) spiked with B were also analysed.
Results: Sensitivity of SBEM was 95%. Selectivity of SBEM was excellent with bacterial growth suppression of respiratory pathogens in > 90% of cases. B could be recovered from SBEM even when other respiratory pathogens were present in 1000x higher concentration. SBEM allowed detection of B from all sputum samples studied even at low level.
Conclusion: SBEM allows selective enrichment of B and suppresses the growth of other respiratory pathogens. Because this medium is simple to prepare with widely available reagents, it can be universally used in clinical laboratories.

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Immunological diagnosis of chronic *Pseudomonas aeruginosa* lung infection in cystic fibrosis patients: a comparison of methods

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Several serological assays determining antibodies of different classes against specific antigens of *P. aeruginosa* (PA) have been found to be value in monitoring course of infection, but the clinical relevance of these tests is not fully understood. The following methods: precipitating antibodies against PA whole-cell sonic extracts by means of crossed-immunoelectrophoresis, IgG and IgA titers against PA lipopolysaccaride (ELISA), and antibodies against PA alkaline protease (AP), elastase (ELA), exotoxin A (EXO A) determined by ELISA have been tested on sera of CF 36 patients. 23 out of 36 patients were chronically colonized and 13 patients were not colonized by PA.
Precipitins and antibodies against PA virulence factors show the higher specificity and positive predictive value (100%), while sensitivity of the two tests is rather low (78 and 52). ELISA test for specific IgG and IgA shows higher sensitivity (96%) however the presence of IgA can not be used for diagnostic purpose, due to low speficity (38%).
In conclusion, none of the tests is able to detect the antibody response in all PA-infected patients. Therefore the use of at least two different methods should be recommended.

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Pilot study to detect the presence of transmissible *Pseudomonas aeruginosa* (PsA) strains in cystic fibrosis (CF) patients attending Birmingham Children's Hospital

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Background: A recent study[1] looking at prevalence of transmissible PsA strains in England & Wales identified the Midlands 1 strain as the second most common genotype, present in 10% of the study population. The study population comprised of adults, some of whom would have previously attended our institution, which is the tertiary paediatric CF centre in West Midlands. Therefore as part of an ongoing study genotyping PsA from all our patients, we decided to look for the presence of the Midlands 1 strain in our isolates.
Methods: 6 PsA isolates identified as the Midlands 1 strain were kindly supplied by Dr F W Scott, HPA, Colindale, London.25 PsA isolates were obtained from children (0-16 yrs) attending our institution and their identity confirmed by biochemical profiles. Genotyping was carried out by pulsed field gel electrophoresis (PFGE) following macrorestriction of chromosomal DNA with the restriction endonuclease *Xba*I.
Results: PsA isolates from our 25 patients were distinguishable by PFGE and none showed strain relatedness to the Midlands 1 strain.
Conclusion: The transmissible Midlands 1 strain was not isolated from any of the paediatric population in the West Midlands tested to date at Birmingham Children's Hospital.
Reference: FW. Scott, TL. Pitt. Identification and characterization of transmissible *Pseudomonas aeruginosa* strains in cystic fibrosis patients in England and Wales. J Med Microbiol 53 (2004), 609-615.

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Does sputum induction (SI) increase recognition of an epidemic strain of *Pseudomonas aeruginosa* (PA)?

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Aim To determine whether sputum induction (SI) increases recognition of the Liverpool Epidemic Strain (LES) of PA in children with cystic fibrosis (CF).
Methods Routine respiratory culture was obtained (sputum or cough swab) followed by a standardized SI protocol with 3% hypertonic saline nebulised for 4 three min periods. Gargle and mouth wash samples were collected before and after SI.
Results 36 children were recruited (10 without CF). In 11 CF children (chronically infected with LES and productive of sputum), LES was grown from both sputum and SI samples. LES was identified in pre-SI gargle or mouth wash samples in 7/11 (and in post samples from 11). Of 9 CF children chronically infected with PA but not normally productive of sputum (determined by physiotherapy assessment), one grew LES in the SI sample and from a preceding cough swab. After SI, that patient grew LES in their mouthwash and gargle. Six CF children had not grown PA from respiratory cultures for at least 12 months and did not grow LES. One child without CF grew LES in their SI sample (light growth). That child had traveled for over an hour in a car with his friend, who has CF and is chronically infected with LES and productive.
Conclusions There is no evidence from this study that SI offers an advantage over standard respiratory culture in the identification of LES, however the small number of positive samples in the non-productive group makes interpretation of these data difficult. In addition this study gives some insight into the biology of this widespread epidemic strain of PA, in particular with apparent transmission following a car journey.